

X-RAY DIFFRACTION STUDIES OF PENICILLIN
AND RELATED COMPOUNDS

BY
KENNETH JAMES PIPENBERG

B. S., Wheaton College, 1943

AN ABSTRACT OF A THESIS

SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS
FOR THE DEGREE OF DOCTOR OF PHILOSOPHY IN CHEMISTRY
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UNIVERSITY OF ILLINOIS, 1946

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INTRODUCTION

In the Fall of 1928, Dr. Alexander Fleming of the University of London discovered that a greenish mold of the *penicillium* variety produced a substance which is capable of destroying certain gram-positive organisms such as staphylococcus, streptococcus, and pneumococcus. The substance derived was named *penicillin* by Fleming. An article describing his findings appeared in 1929, but little more was done with this new knowledge. Some ten years later Dr. Howard Florey of Oxford University reinvestigated this strange substance with the result that penicillin was introduced to the medical profession as a chemical therapeutic. The demands of war-time medicine hastened the development greatly. The results of Florey and co-workers led to an expansion of the program under the Rockefeller Foundation and the British Medical Research Council.

Because of the incessant bombing of England in the Spring of 1941, a large share of the research and development on the drug was transferred to the United States, where a vast program under the Office of Scientific Research and Development was instituted. The production of penicillin mounted from very meager yields of two units per cubic centimeter to two hundred units per cubic centimeter. With this, the manufacture became feasible on a commercial scale, and American large-scale production took charge. The supply of penicillin moved first to the military forces and then as production increased, the civilian needs were supplied.

As a part of the Penicillin Project x-ray diffraction studies of penicillin and related compounds were carried on in this laboratory (1). All the samples used in these studies were supplied through the kind cooperation of the various research groups connected with the Committee on Medical Research. Each of the samples received has been examined by powder

diffraction methods, and wherever the sample was in the form of single crystals of suitable size, oscillation, rotation, and Weissenberg technics were applied to determine the unit cell dimensions and the space group. The experimental methods very conveniently divide the studies into two main topics—identification and differentiation from powder diffraction patterns and crystallographic data obtained from single crystal analysis.

IDENTIFICATION AND DIFFERENTIATION FROM POWDER

DIFFRACTION PATTERNS

In the course of this investigation fifty-two samples were studied by powder diffraction methods. The types of compounds represented were sodium penicillin G, sodium penicillin F, clinical sodium penicillin, sodium penicillin X, ammonium penicillins, methyl penicillin G, thiocyanate derivatives of methyl penicillin G, esters of penicilloic acid, synthetic compounds with lactam ring structures, penillic acid, sulfone derivatives of methyl penicillin G, and a variety of miscellaneous compounds.

1. The samples of sodium penicillin G gave no evidence for polymorphism or hydration.

2. Each sample of sodium penicillin F gave a different diffraction pattern (2). The existence of polymorphism and solvation is strongly indicated.

3. Studies of clinical penicillin were made in the hope that the decomposition which occurs on standing could be detected and the products identified. No preceptable change was noted over a period of thirteen months; this does not indicate that decomposition did not occur, but rather, the method did not detect the decomposition.

4. Sodium penicillin X could be distinguished from the F and G varieties.

5. The ammonium penicillins (3) provided the problems of solvation and polymorphism, and in addition, the problem of complex mixtures of the various types of penicillin.

6. Only one form of methyl penicillin G could be found. Its diffraction pattern is very distinct from that of sodium penicillin G.

7. A series of thiocyanate derivatives of methyl penicillin G were studied. No methyl penicillin G was found present in the derivatives. Upon treatment with alkali the original derivative of the methyl ester gave a compound having a new diffraction pattern; upon treatment with hydrochloric acid the original derivative gave a compound having still another diffraction pattern. The compound obtained by alkali-treatment was further treated with hydrochloric acid, the resulting compound gave a diffraction pattern identical with the compound obtained by the hydrochloric acid treatment of the original derivative. A synthetic compound was examined which was identical with the hydrochloric acid hydrolysis product. A compound, prepared from the original thiocyanate derivative and believed to be a diastereoisomer of the alkali-treated product, was found to be identical with another synthetic compound which had been examined. The results indicate that the alkali-treatment produces an isomerization product, while the hydrochloric acid treatment produces a hydrolysis product. Possible structures (4) for these compounds are discussed and the most probable are suggested.

8. A variety of esters of penicilloic acid were examined. No distinction could be made between the optical isomers of the α -methyl ester. The α -methyl, α -ethyl, and α -ethyl β -methyl esters were found isomorphous with each other, while the α , β -dimethyl ester was strikingly different.

9. Synthetic compounds having a δ -lactam ring structure were compared with sodium and methyl penicillin G. The compounds would be the next higher homolog of penicillin G, if penicillin G has a β -lactam ring structure. The free acid and the methyl ester of the δ -lactam acid gave patterns very similar to methyl penicillin G but very unlike sodium penicillin G.

10. Penillic acid G was identified and an unknown penillic acid was examined. The unknown compound is probably a mixture of the F and K acids. This is substantiated by the analytical data.

11. A sulfone and a hydrogenated sulfone of methyl penicillin G were compared with pure methyl penicillin G. The expected expansions in the lattice dimensions were observed.

12. A number of miscellaneous compounds were studied. It was thought these compounds would aid in finding structural similarities with some of the penicillins or their derivatives, but the studies have shown them otherwise.

CRYSTALLOGRAPHIC DATA FROM SINGLE CRYSTAL ANALYSIS

The most popular single crystal methods—Laue, rotation, oscillation, and Weissenberg—are considered briefly. As an aid in interpreting the rotation and Weissenberg patterns, the reciprocal lattice concept is introduced.

Making use of the single crystal methods, three compounds were studied. A fourth compound was analyzed by the application of the reciprocal lattice method to a powder pattern.

1. Methyl penicillin G was found to have an orthorhombic cell with unit dimensions $a_0=40.92$ A., $b_0=7.75$ A., and $c_0=17.27$ A. The density is 1.25 g/cm³.; the molecular weight is 348.4; and the number of molecules per unit cell is twelve.

2. α -Ethyl β -methyl penicilloate G has an orthorhombic cell with the unit dimensions $a_0=20.10$ A., $b_0=18.10$ A., and $c_0=5.41$ A. The density is 1.33 g/cm³.; the molecular weight is 394; the number of molecules per unit cell is four; and the space group is $P 2_1 2_1 -$.

3. The methyl ester of the acid having a β -lactam ring structure has a tetragonal cell with the dimensions $a_0=11.01$ A., and $c_0=15.90$ A. The space group is $P4/m$.

4. The free acid having a β -lactam ring structure was not obtainable as a single crystal, but the powder pattern yielded the data from reciprocal lattice projections. The crystal system is tetragonal. The cell dimensions are $a_0=10.42$ A., and $c_0=15.31$ A. Since the free acid is isomorphous with the methyl ester, the space group is $P4/m$ also.

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VITA

The author was born September 20, 1920, in Racine, Wisconsin, and received his primary and secondary education in the public schools there. After graduation from Washington Park High School in 1939, he entered the University of Wisconsin Extension Division in Milwaukee. In September, 1940, he transferred to Wheaton College, and in June, 1943, received the degree of Bachelor of Science in Chemistry. He entered the Graduate School of the University of Illinois in the Fall of 1943. While at Illinois, he held a teaching assistantship in the Analytical Division of the Department of Chemistry from October, 1943, through February, 1945; a special research assistantship under contract OEMcmr-439 from March, 1945, through December, 1945; and the Continental Oil Fellowship from January, 1946, through June, 1946.

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